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SEX DIFFERENTIATION IN LARVAL INSECTS.

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The question of the causes of sex differentiation is a problem to the solution of which we seem now to be only a little nearer, despite numerous researches, than we were many years ago. It is advisable, perhaps, to continue to attempt to overcome some of the outworks of this well entrenched problem. One of the outliers of the main problem may be described in the phrase "When is sex differentiated?", another in the phrase "Does nutrition affect sex?"

Being engaged in rearing experimentally large numbers of silkworm moths, *Bombyx mori*, I have taken advantage of the

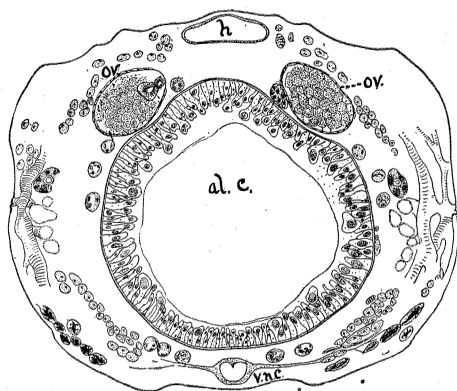


FIG. 1. Section through female *Bombyx* larva, just after the third moult; *h*, heart; *al. c.*, alimentary canal; *v. n. c.*, ventral nerve cord.

opportunity to test for this species both these subsidiary parts of the sex differentiation problem. Various lots of larvæ were set apart, each individual being isolated so as to insure identity of nutrition conditions, and fed on short rations. The result of these experiments is given in a paper in the *Journal of Experimental Zoology* (vol. I, pp. 357-360, 1904). It is sufficient

to say here, in a word, that this "short feeding" produced no apparent effect in determining the sex of the moths.

Since these experiments I have learned a good reason why

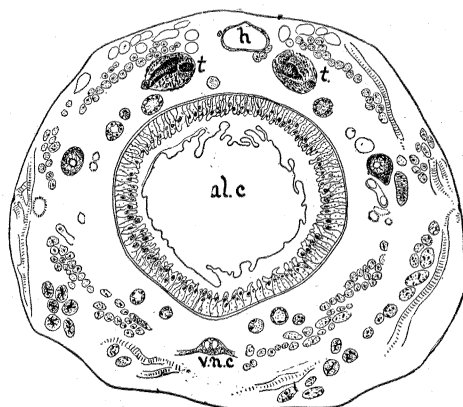


FIG. 2. Section through male *Bombyx* larva, just before the third moult; *h*, heart; *a.l.c.*, alimentary canal; *v. n. c.*, ventral nerve cord; *t, t*, testes.

the nutrition of the larva, at least after its first moulting, should have no effect on the sex as revealed in the moth, and this reason is that the sex of each individual is definitively determined at

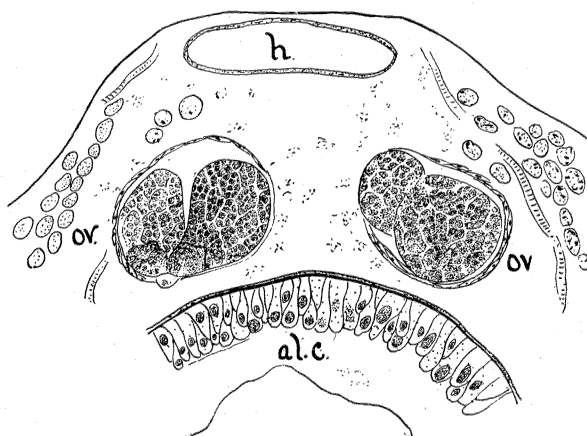


FIG. 3. Section (dorsal third) through female *Bombyx* larva, just after the second moult; *h*, heart; *a.l.c.*, alimentary canal; *ov, ov*, ovaries.

least as early as immediately after the first larval moulting, as may be readily perceived by an examination of the rudimentary reproductive glands.

In the larval silkworm there may be noted on the dorsal wall of the fifth abdominal segment two low tubercles, rather dark colored. Directly beneath these spots lie the developing reproductive organs (ovaries, testes). By dissecting a number of

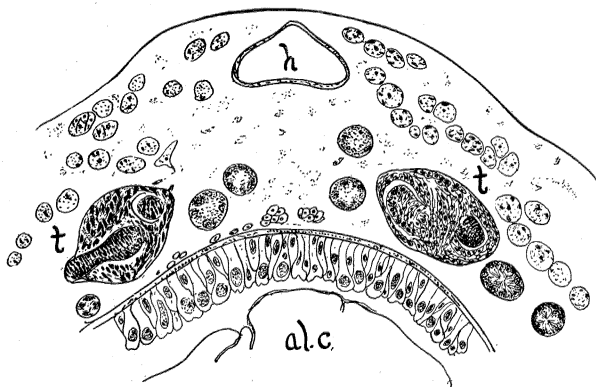


FIG. 4. Section (dorsal third) through male *Bombyx* larva, just after the second moult; *h*, heart; *al. c.*, alimentary canal; *t*, *t*, testes.

larvæ in their last or next to last intermoult period, a marked difference will be noted in the size of the organs in different individuals. By dissecting out the organs and sectioning them,

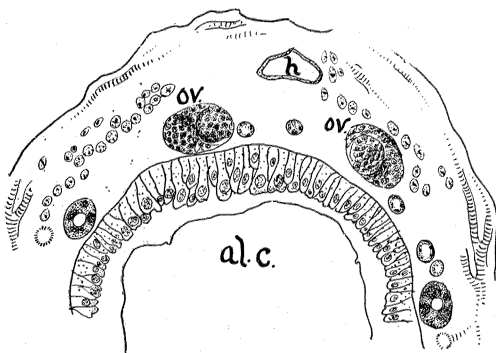


FIG. 5. Section (dorsal half) through female *Bombyx* larva, just after the first moult; *h*, heart; *al. c.*, alimentary canal; *ov*, *ov*, ovaries.

constant marked histologic differences will be seen in the two sizes of organs. As a matter of fact the larger are ovaries, with well-developed rudiments of egg-tubes, the larvæ possessing

them being female individuals, while the smaller are testes. By dissecting larvæ of successively younger age, these differences in size and histologic character may be followed back to just after the first moulting.

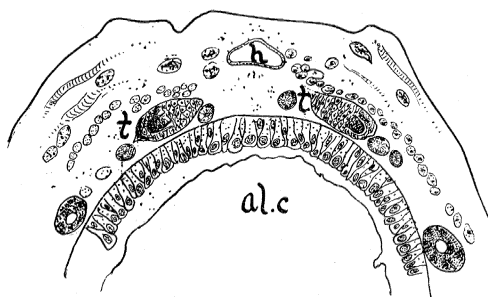


FIG. 6. Section (dorsal half) through male *Bombyx* larva, just after the first moult; *h*, heart; *al. c*, alimentary canal; *t*, *t*, testes.

Figs. 1 to 8 show, with little need of explanatory description, the gross character of the histologic differences, the relative sizes (in cross-section) of the two kinds of organs, and also the rela-

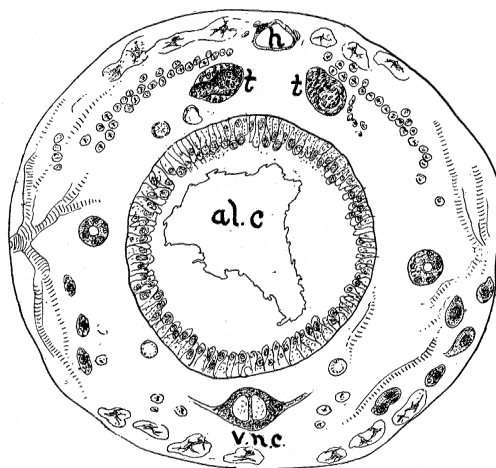


FIG. 7. Section through male *Bombyx* larva, just before the first moult; *h*, heart; *al. c*, alimentary canal; *v. n. c*, ventral nerve cord; *t*, *t*, testes.

tive size of these organs in the various larval stages. I have not been sure of being able to distinguish between the sizes or the histologic characters of the reproductive organs in the just

hatched larvæ, but even here the organs are conspicuous and well started in development, being larger in diameter than the heart or the spinning glands.

We may affirm then positively that from the time of the first moulting on the silkworm larva has its sex determined: its reproductive glands are ovaries or testes distinguishable by obvious histologic characteristics. Also that the just hatched larva has reproductive organs already well developed. I have little doubt that careful scrutiny of the organs at this stage would reveal to the trained histologist, especially to the student of oögenesis

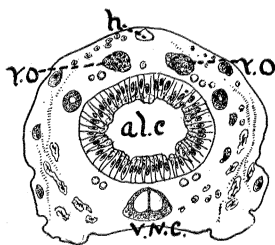


FIG. 8. Section through *Bombyx* larva (male or female?), two days after hatching; *h*, heart; *al. c*, alimentary canal; *v. n. c*, ventral nerve cord; *r. o*, reproductive organs.

and spermatogenesis, definitive characteristics marking sex differentiation. The sex of the silkworm is not to be tampered with by gorging or starving, and what is true of this lepidopteron is undoubtedly true of its cousins, the other moths and the butterflies. It is probably also true of other insects with complete metamorphosis. I recall dissections of various larvæ, notably of *Corydalis cornuta* (a neuropteran) and of *Holorusia rubiginosa* (a dipteran) in which the reproductive organs appear of two sizes in specimens of the same age: indeed in *Corydalis*, of two shapes. These organs need histologic examination. Some student should laboriously work through a long and representative series of insects and settle the question as to the time of sex differentiation. That is, find out whether it be true for all, as it is in the silkworm, that the time of sex differentiation is obvious before, or, at latest, at very little after the time of hatching. If it is true, the question of the influence of nutrition in sex determination will also be settled — for insects. And we need waste no more time in tedious feeding and tabulating.